

# Urine Based Assays for Bladder Cancer

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APMG

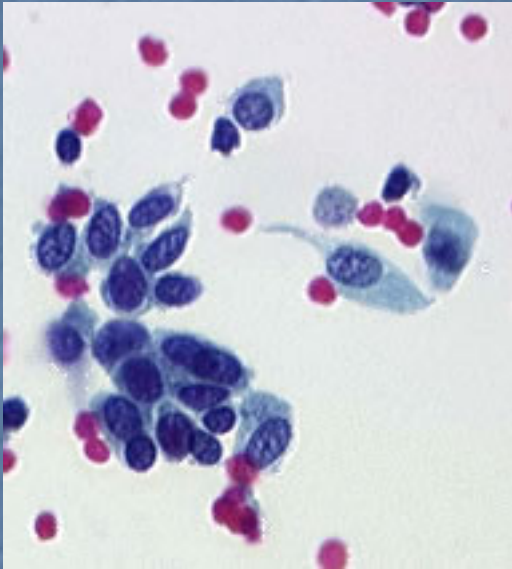
# Background

- 57,000 annual new cases of bladder cancer in US
- 12,500 will die from disease
- M>F
- Fourth most common cancer in men  
Ninth most common in women
- Caucasians>African Americans

# Bladder Cancer Risk Factors

- Spinal cord injury
- Cigarette smoking
- Aniline dyes

# Laboratory



- Hematuria most common
  - >90% of hematuria patients do not have bladder cancer
- Urine cytology limited
  - Observer variability
  - Few cells
  - Poor preservation
  - Sensitivity low, specificity high (>98%)

# Urine Bladder Cancer Markers

- Cytology based
- Urine based



# Cytology Based

- Fluorescence-in-situ-hybridization (FISH)
  - 14 urinary specimens labeled with different DNA labeled fluorochromes directed against the chromosomes 3,7, and 17, and a locus specific probe against 9p21
- Results

11/14 specimens were evaluable

Of 6 cases with 22 or less abnormal cells, 4 were scored as atypical by cytology and 2 as UC

All 5 cases with >2 abnormal cells were high grade UC

Two cases of high grade UC showed 90% homozygous deletion for 9p21, while the remaining cases showed a wide range of LOH for 9p21

- Conclusion
  - Multi-color FISH probe test more sensitive than cytology

# Urine Based Assays

- Bladder tumor antigen (BTA)
- NMP22
- Fibrinogen degradation protein (FDP)

# Bladder Tumor Antigen (BTA)



- Quantitative Automated and stat physician office test
- Identifies a complement-related factor H molecule in urine of bladder cancer patients with bladder cancer
- Sensitivities (24% to 89%)  
Specificity (26% to 93%)
- Increase in BTA with hematuria
- Poor sensitivity and specificity limits use



# NMP-22 (Matrictech)



- Quantitative microplate enzyme immunoassay performed on single voided urine sample
- Nuclear Matrix Protein found in human epithelial cells
  - Nuclear protein known as NuMa, protein component of the mitotic apparatus which is found in all eukaryotic cells
  - Bladder cancer release large quantities of NMP22
- Negative Predictive Value of >86%  
Sensitivity (38% to 100%)  
Specificity (61% to 95%)
- Unaffected by gross hematuria and common intravesical treatments such as BCG or thiotepa

# Fibrinogen degradation protein (FDP)

- Sensitivities (76% to 81%)  
Specificities (75% to 86%)
- Taken off market and not currently available in United States.

# Urine Assay Types

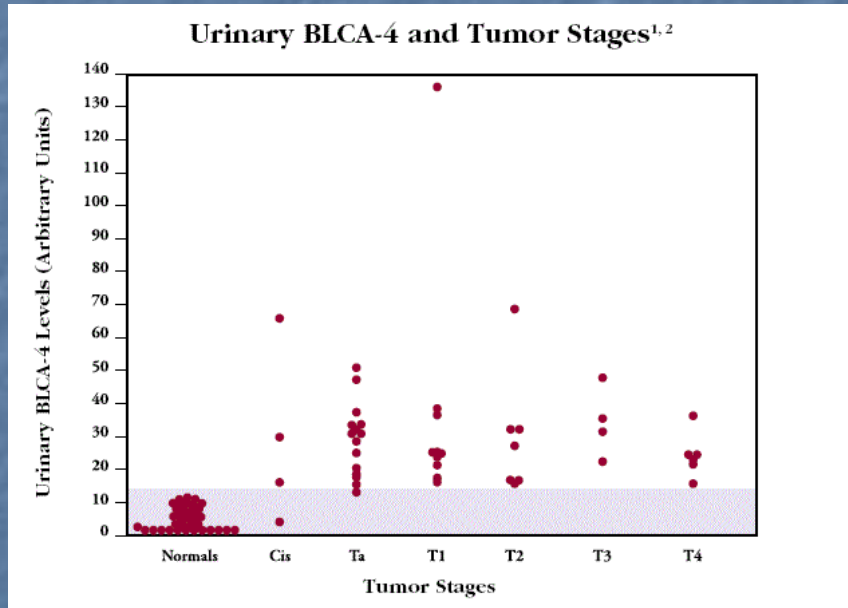
- Detect nucleic acids or nucleic acid alterations within the urine
  - Microsatellite alterations examine loss of heterozygosity or instability
- Detect Enzyme
  - Telomerase in the urine of patients with bladder cancer
  - hTert mRNA determined by RT-PCR, sensitivity (62% to 85%) specificity (60% to 96%)
  - 265 patients, 55% of which had recurrent disease
    - Sensitivity 46% significantly lower than the other assays

# Markers In Development

- UBC detects cytokeratin 8 and 18
  - Sensitivities (72.3% to 86.7%)  
Specificities (71.8% to 86.5%)
- CYRFA-21-1 detects cytokeratin 19
  - Sensitivity (83% to 100%) and a specificity (74% to 100%)
- RT-PCR Cytokeratin 20 mRNA levels
  - Sensitivity (82% to 91%) specificity (67% and 97%)
- Lewis X antigen
  - Sensitivities (80% to 81%) and a specificity (86%)
- Hyaluronidase and hyaluronic acid assays
  - Detect GAG-linked proteins specific for bladder cancer
  - Sensitivities (90% to 92%)  
Specificities (84% to 92%)
- Survivin
  - Inhibitor of apoptosis
  - Sensitivity of 100% but the specificity not determined



# BLCA-4 (Eichrom Technologies)



- Present in people with bladder cancer including both tumor and normal regions (field effect)
  - Not found in the bladder of individuals without the disease
- Transcriptional regulator of gene expression for bladder cancer
- Immunoassay on straight urine samples
  - Clinical trial of 106 individuals  
Sensitivity of 96.4% and a specificity of 100%, compared to cytology
- Spinal cord patients
  - Specificity high, not elevated with cystitis, smoking
  - Identified asymptomatic bladder cancer patients
- National clinical trials underway



# References

- Getzenberg RH. Laboratory Medicine  
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